

< The purpose of >  
 << Wherein said assay is >>

# CLAIMS

Use of red blood cells in an

1. ~~An assay for detecting type IV cytosolic phospholipase A<sub>2</sub> (cPLA<sub>2</sub>) or a protein immunologically homologous to type IV cPLA<sub>2</sub>, the assay comprising use of red blood cells~~

2. ~~An assay~~ according to claim 1 <sup>Use</sup> for use in the diagnosis of a disease in which dysfunction of cell signalling systems involving highly unsaturated fatty acids is implicated.

3. ~~An assay~~ according to claim 1 <sup>Use</sup> for use in monitoring the effectiveness of medication administered to a patient suffering from a disease in which dysfunction of cell signalling systems involving highly unsaturated fatty acids is implicated

4. ~~An assay~~ according to claim 1 <sup>Use</sup> for use in drug development for a disease in which dysfunction of cell signalling systems involving highly unsaturated fatty acids is implicated

5. A method of diagnosis of a disease in which dysfunction of cell signalling systems involving highly unsaturated fatty acids is implicated, said method comprising the detection of type IV cytosolic phospholipase A<sub>2</sub> (cPLA<sub>2</sub>) protein or a protein immunologically homologous to type IV cPLA<sub>2</sub> in or on red blood cells.

6. A method of monitoring the effectiveness of medication administered to a patient suffering from a disease in which dysfunction of cell signalling systems involving highly unsaturated fatty acids is implicated, said method comprising the detection of type IV cytosolic phospholipase A<sub>2</sub> (cPLA<sub>2</sub>) protein or a protein immunologically homologous to type IV cPLA<sub>2</sub> in or on red blood cells.

7. A method of drug development for a disease in which dysfunction of cell signalling systems involving highly unsaturated fatty acids is implicated, said method comprising the detection of type IV cytosolic phospholipase A<sub>2</sub> (cPLA<sub>2</sub>) protein or a protein immunologically homologous to type IV cPLA<sub>2</sub> in or on red blood cells.

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8. ~~An assay~~ <sup>Use</sup> or method according to any one of claims 1 to 7 wherein the red blood cells are isolated from the human body.

5 9. ~~An assay~~ <sup>Use</sup> or method according to any one of claims 1 to 7 wherein said assay or method comprises use of a whole blood sample without prior isolation of said red blood cells.

10 10. ~~An assay~~ <sup>Use</sup> or method according to any one of claims 2 to 9 wherein said disease is a disease or disease process in which type IV cPLA<sub>2</sub> activity or concentration is altered from normal levels.

15 11. ~~An assay~~ <sup>Use</sup> or method according to anyone of claims 2 to 9 wherein said disease is a disease or disease process in which type IV cPLA<sub>2</sub> activity or concentration is increased.

20 12. ~~An assay~~ <sup>Use</sup> or method according to any one of claims 2 to 11 wherein the disease is schizophrenia, dyslexia, bipolar or manic depressive illness, cachexia or brain injury.

25 13. ~~An assay~~ <sup>Use</sup> or method according to claim 12 wherein the brain injury is stroke or mechanical brain injury.

30 14. ~~An assay~~ <sup>Use</sup> or method according to any one of claims 1 to 13 wherein the type IV cPLA<sub>2</sub> protein or the protein immunologically homologous to type IV cPLA<sub>2</sub> has a molecular weight in the range 80 to 110 kDa or in the range 70 to 80 kDa or in the range 50 to 60 kDa.

30 15. ~~An assay~~ <sup>Use</sup> or method according to any one of claims 1 to 13 wherein the type IV cPLA<sub>2</sub> protein or the protein immunologically homologous to type IV cPLA<sub>2</sub> has a molecular weight in the range 90 to 105 kDa or in the range 70 to 80 kDa or in the range 50 to 60 kDa.

<Wherein said assay or method comprises>

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Use

16. ~~An assay~~ or method according to any preceding claim <sup><></sup> comprising the steps of collecting a sample of blood from a subject and detecting the proteins *ex vivo*.

Use

17. ~~An assay~~ or method according to claim 16 <sup><></sup> further comprising one or more of the steps of separating the red cells from the other blood components, disrupting the red cells, detecting the proteins either directly or following a protein separation technique.

Use

18. ~~An assay~~ or method according to claim 17 wherein the red cells are disrupted by sonication, freezing, nitrogen cavitation or lysis.

Use

19. ~~An assay~~ or method according to any preceding claim wherein said proteins are detected by immunoassay.

Use

20. ~~An assay~~ or method according to any preceding claim wherein said proteins are detected using an antibody or antibodies that recognise an epitope or epitopes from amino acids 82 to 749 of type IV cPLA<sub>2</sub> protein from human monocyte (U937) cells.

Use

21. ~~An assay~~ or method according to any preceding claim wherein said proteins are detected using an antibody or antibodies raised against an epitope or epitopes from amino acids 82 to 749 of type IV cPLA<sub>2</sub> protein from human monocyte (U937) cells or raised against an epitope or epitopes of a synthetic peptide matching amino acids 82 to 749 of type IV cPLA<sub>2</sub> protein from human monocyte (U937) cells.

Use

22. ~~An assay~~ or method according to claim 20 or 21 wherein said epitope or epitopes are from a peptide sequence or sequences which comprise the catalytic centre of type IV cPLA<sub>2</sub> protein from human monocyte (U937) cells.

Use

23. ~~An assay~~ or method according to claim 20 or 21 wherein said epitope or epitopes are from the peptide sequence of amino acids 241 to 260 of type IV cPLA<sub>2</sub> protein from human monocyte (U937) cells.

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*Use*

24. ~~An assay~~ or method according to claim 19 wherein said proteins are detected using an antibody or antibodies raised against an epitope or epitopes from amino acids 1 to 216 of type IV cPLA<sub>2</sub> protein from human monocyte (U937) cells.

*Use*

25. ~~An assay~~ or method according to claims 20, 21, 22, 23 or 24 wherein two or more of the antibodies are used in combination or in sequence to detect the said proteins with the required specificity.

*Use*

26. ~~An assay~~ or method according to any of claims 1 to 18 for detecting type IV cPLA<sub>2</sub> wherein said proteins are detected by substrate assay.

*other than type IV cPLA<sub>2</sub>*

27. A protein obtainable by isolation from red blood cells, said protein being immunologically homologous to type IV cPLA<sub>2</sub> and having a molecular weight in the range 80 to 110 kDa or a molecular weight in the range 70 to 80 kDa or a molecular weight in the range 50 to 60 kDa.

28. A protein according to claim 27, said protein being immunologically homologous to type IV cPLA<sub>2</sub> and having a molecular weight in the range 90 to 105 kDa or a molecular weight in the range 70 to 80 kDa or a molecular weight in the range 50 to 60 kDa.

29. A diagnostic kit comprising means for disrupting red blood cells and further comprising an antibody or antibodies to a protein obtainable by isolation from red blood cells, said protein being type IV cPLA<sub>2</sub> protein or a protein immunologically homologous to type IV cPLA<sub>2</sub>.

30. A diagnostic kit according to claim <sup>29</sup>~~28~~ wherein said antibody or antibodies is/are raised against an epitope or epitopes from amino acids 82 to 749 of type IV cPLA<sub>2</sub> protein from human monocyte (U937) cells.

31. A diagnostic kit according to claim <sup>29</sup>~~28~~ wherein said antibody or antibodies is/are raised against an epitope or epitopes from a peptide sequence or sequences which

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comprise the catalytic active centre of type IV cPLA<sub>2</sub> protein from human monocyte (U937) cells.

24, 30 or 31

32. A diagnostic kit according to claim ~~28, 29 or 30~~ wherein said means for disrupting red blood cells is a means for lysing red blood cells.

24, 30 or 31

33. A diagnostic kit according to claim ~~28, 29 or 30~~ which is suitable for near-patient testing.

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